WASTE WATER AS RESERVOIR OF ANTIBIOTIC RESISTANT MICRO-ORGANISMS:

A CASE OF LUANSHYA WASTE WATER PONDS

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ABSTRACT

Background and objectives: The greatest threat to the use of antibiotics in the treatment of bacterial infections is the emergence and spread of antibiotic resistant bacteria. Indiscriminate use of antibiotics for medical purposes has taken the brunt of the blame; in addition patients who do not complete their prescriptions have also been recognized as a cause for the development of antibiotic resistant bacteria. Antibiotic resistant microorganisms get to the environment through different means amongst them the pollution of the environment by human beings. Pollution of the environment by human and animal waste is a common threat to water resources. Wastewater discharged in rivers may increase the prevalence of multidrug resistant microorganisms in natural waters. Untreated human sewerage frequently enters water ways as a point of discharge or by accident from waste water treatment plants. The main objective of this study was to determine the presence of antibiotic resistant microorganisms in Luanshya waste water ponds using the disc diffusion method.

Materials and Methods: Samples were collected from the waste water ponds in Luanshya using the standard methods and then pooled into nutrient broth before incubating at 37 degrees for 24 hrs. On the following day colonies were sub cultured to make pure cultures by touching a single colony and inoculating on media. The microorganisms from pure cultures were identified on the basis of their colony morphology, appearance on MacConkey agar and subsequent biochemical tests.

Results: It was observed that coliforms that were isolated from the waste water ponds in Luanshya were resistant to one or more drugs they were subjected to and E.coli was the most resistant organism isolated.

Conclusions: In order to reduce the spread of antibiotic resistant microorganisms there is need for the reduction of waste water discharge in aquatic environment, waste water treatment plants should treat the sewage adequately before discharging into the receiving waters. Use of night soil as manure and irrigating of crops with waste water should be discouraged or stopped all together.

Key words: Waste water ponds, coliform, antibiotic discs, Susceptibility testing, Kirby-bauer

1.0 INTRODUCTION

The greatest threat to the use of antibiotics in the treatment of bacterial infections is the emergence and spread of antibiotic resistant bacteria that consequently cannot be treated by previously successful regimes [1].

In a study carried out by Katherine et al., 2007 the researcher stressed the need to characterize the occurrence, fate and transport of both the antibiotics and the antibiotic resistant microorganisms [2]. They reported that antibiotic resistant microorganisms get to the environment through different means amongst them the pollution of the environment by human beings.

Pollution of the environment by human and animal waste is a common threat to water resources. Wastewater discharged in rivers may increase the prevalence of multidrug resistant microorganisms in natural waters. Untreated human sewerage frequently enters water ways as a point of discharge or by accident from waste water treatment plants [3, 4]. Khachatourians in 1998 reported that, resistant bacteria may transfer their resistance genes to
previously non-resistant pathogens bacteria or directly infect human beings [5]. Similarly Bridget et al, in 2010 observed that there was a frequent conjugal transfer of resistant genes in microorganisms that were assayed [6]. They observed that 83.3% of bacteria isolated from the environment which had resistance genes were able to transfer one or more of their resistant genes to strains of *Salmonella typhimurium*. They also observed that 71.35% of faecal coliforms which had resistant genes were able produced viable transconjugates, and 27.4% were able to transfer all their resistant determinants to other coliforms.

Although antibiotic resistance has involved extensive research in clinically relevant human pathogens, environmental Reservoir of antibiotic resistant determinants and their contribution to resistance in clinical settings have only been considered in the last decade [7, 8].

Indiscriminate use of antibiotics for medical purposes has taken the brunt of the blame, namely by those physicians who prescribe antibiotics against viral disease, (Bolaji et al, 2011) and that people who do not finish their prescription drugs may have caused the increase of antibiotic resistant microorganisms. Studies on antibiotic resistance in wastewater have been done in high income countries, while studies in low income countries are few and sparsely[9].

This study was primarily carried out to investigate the presence of antibiotic resistant bacteria in waste water ponds in Luashya.

2.0 MATERIALS AND METHODS

2.1 Sampling sites

Samples were collected from Luanshya wastewater ponds. Luanshya is a town in Zambia located on the Copperbelt province. Below is the map of the town and satellite view of the waste water ponds.
2.2 Sampling, collections, and bacteriological analysis

Waste water samples were collected in sterile 50ml sample collecting bottles using a sampling container. The samples were collected in the morning and transported to the laboratory within one hour of collection. Five samples from Luanshya waste water ponds were collected. (Two samples were collected in the second week). The samples were transferred and pooled into a bottle containing nutrient broth and incubated overnight at 37 degrees. Thereafter a Swab was used to inoculate on blood and MaCconkey agar and incubated for 24 hours at 37 degrees. On the following day colonies were sub cultured to make pure cultures by touching a single colony and inoculating on media. The microorganisms from pure cultures were identified on the basis of their colony morphology, appearance on MaCconkey agar and subsequent biochemical tests.

2.3 Antibiotic susceptibility testing

Antibiotic susceptibility of bacterial isolates was assayed according to Kirby-Bauer disc diffusion [10]. All the media plates were put in the incubator for ten minutes inoculation and placement of antibiotic disc to allow excess moisture to dry from the media plates. Susceptibility test results were interpreted according to performance standards for antimicrobial disc susceptibility tests, M100.S22, CLSI vol 32 no3, January 2012.

2.3.1 Antibiotic discs used


2.0 RESULTS

Table 1 Microorganisms and Drugs they are resistant to

<table>
<thead>
<tr>
<th>E.coli</th>
<th>S.pneumonia</th>
<th>Pseudomonas.sp</th>
<th>Enterobacteria.sp</th>
<th>Proteus.sp</th>
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</thead>
<tbody>
<tr>
<td>Cotrimoxazole</td>
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<tr>
<td>Tetracycline</td>
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<td>Amoxylin</td>
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<td>Nalidixic Acid</td>
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<td>Kanamycin</td>
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<td>Penicillin</td>
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<td>Nitrofurantoin</td>
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<td>Chloramphenicol</td>
<td>x</td>
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Key: X-the drug to which the microorganism was observed to be resistant using the disc diffusion method.

Note that only the drugs that the isolated bacteria was resistant to have been shown in the table.

3.0 DISCUSSION

The presence of antibiotic resistant bacteria in an environment maybe an indication, that the area is contaminated with antibiotics [11]. In waste water, these antibiotics may come from humans beings. A large part of antibiotics consumed end up in the waste water [12] and the sub lethal concentrations of drugs may exert selective pressure for or maintain Resistance among microorganisms in the waste water. [13, 14].

Antibiotics have been detected in municipal waste water, effluent of waste water treatment plants and the rivers that receive waste water [15, 16]. From the results it was observed that the isolated microorganisms were resistant to at least one drug E.coli was resistant to five drugs. This result may be because E.coli in this study could have been exposed to the tested drugs before. The other explanation would be that E.coli could have acquired these resistant genes from other microorganisms, Bridget et al, 2010 showed that coliforms are able to transfer and receive resistance determinants. The other microorganisms isolated were resistant to fewer drugs in case of Pseudomonas sp three drugs. Enterobacteria sp two drugs, Streptococcus pneumoniae two drugs and Proteus sp one drug [6].

The presence of antibiotic resistant microorganisms calls for concern. The wastewater ponds can be a source of antibiotic resistant diseases. There is need to warn people about the dangers of using wastewater. The findings of this study present a potential health problem as the isolated microorganisms are associated with different diseases [17]. Ahmad et al., 2013 observed that untreated waste water is responsible for contamination of reservoir waters with antibiotics and antibiotic resistant bacteria [15]. The presence of pathogenic antibiotic resistant bacteria in aquatic environments can be a source of antibiotic resistant infections especially if this water is used for drinking, recreational activities or irrigation [18, 19]. Treatment of infection arising from antibiotic resistant organisms is difficult if not impossible [20]. The reuse of waste water raises concern about resistance bacteria budding themselves in the human population. There is need to discourage the use of night soil as fertilizer as well as irrigating of crops using waste water. The increase of urban sewers that may serve as drinking water sources for animals and birds may promote the continuity in the dissemination of antibiotic resistant microorganisms among different animals, birds and human beings populations [21].

5.0 CONCLUSIONS

In order to reduce the spread of antibiotic resistant microorganisms there is need for the reduction of waste water discharge in aquatic environment, waste water treatment plants should treat the sewage adequately before discharging into the receiving waters. Use of night soil as manure and irrigating of crops with waste water should be discouraged or stopped all together.

6.0 ACKNOWLEDGEMENTS

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REFERENCES


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